

# Persistent Pollutants and Factors Affecting Their Accumulation in Rockfishes (*Sebastes* spp.) from Puget Sound, Washington

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## Introduction

As a participant in the Puget Sound Ambient Monitoring Program (PSAMP), the Washington Department of Fish and Wildlife (WDFW) has evaluated contaminant levels in a number of marine and anadromous fish species from 1989 to the present. Among its targets are quillback rockfish (*Sebastes maliger*) and brown rockfish (*S. auriculatus*). These rockfish species are demersal, long-lived<sup>1</sup> carnivores that typically associate closely with complex, high-relief substrates such as rocky reefs (Matthews 1990). Because of their site-fidelity, trophic status, and long life-expectancy, these rockfishes are expected to have a high probability of accumulating persistent pollutants such as mercury (Hg), polychlorinated biphenyls (PCBs), and organochlorine pesticides if present in their environment.

Previous WDFW work (West and O'Neill 1995) identified PCBs and Hg (of 99 organic and inorganic toxic contaminants tested) as the most commonly detected contaminants observed in high enough concentrations to cause concern in rockfish from Puget Sound<sup>2</sup>. That study reported results from 1989–1993 for rockfish muscle tissue composites, where each sample consisted of tissue mixed together from five fish. The compositing procedure is generally used to lower statistical variability in contaminant concentrations to increase the power of detecting differences between locations. However, compositing also dampens the full range of concentrations otherwise observable from individuals. This problem is especially acute for long-lived species like rockfish, where composites may be comprised of fish with a wide range of ages.

In order to obtain a more accurate estimate of the range of individual concentrations of PCBs and Hg in rockfish, as well as to identify more accurately the relationship of these contaminants with factors such as fish age, WDFW initiated analyses of individual rockfish in 1995. The present paper presents results from sampling individual quillback and brown rockfish from 1995 through 1997 for mercury, and 1995–1996 for PCBs.

## Methods

Quillback and brown rockfish were sampled using bottom-trawls and hook-and-line from six locations in the Puget Sound (Figure 1). Rockfish from Double Bluff, San Juan Islands and Blakely Rocks were taken with hook-and-line in October through November, 1995. Rockfish from Sinclair Inlet and Foulweather bluff were taken with bottom trawls in April, 1995 and May, 1997, respectively. Rockfish from Elliott Bay were taken using hook-and-line and bottom trawls in October through November, 1995, and April through May, 1996 and 1997. Sample sizes ranged from eight (for Sinclair Inlet) to 42 (for Elliott Bay); all samples except five brown rockfish from Sinclair Inlet were quillback rockfish. Upon landing, fish were labeled with numbered external tags, measured for total length (mm), wrapped in aluminum foil, sealed individually in plastic bags, and placed on ice for transport to the laboratory. Sagittal otoliths were removed for estimating fish ages; presumed annual increments were counted from broken-and-burned surfaces. In the laboratory, up to 250 g of lateral skeletal muscle tissue was excised from skinned fish within 10 days of collection, and immediately frozen for later chemical analysis.

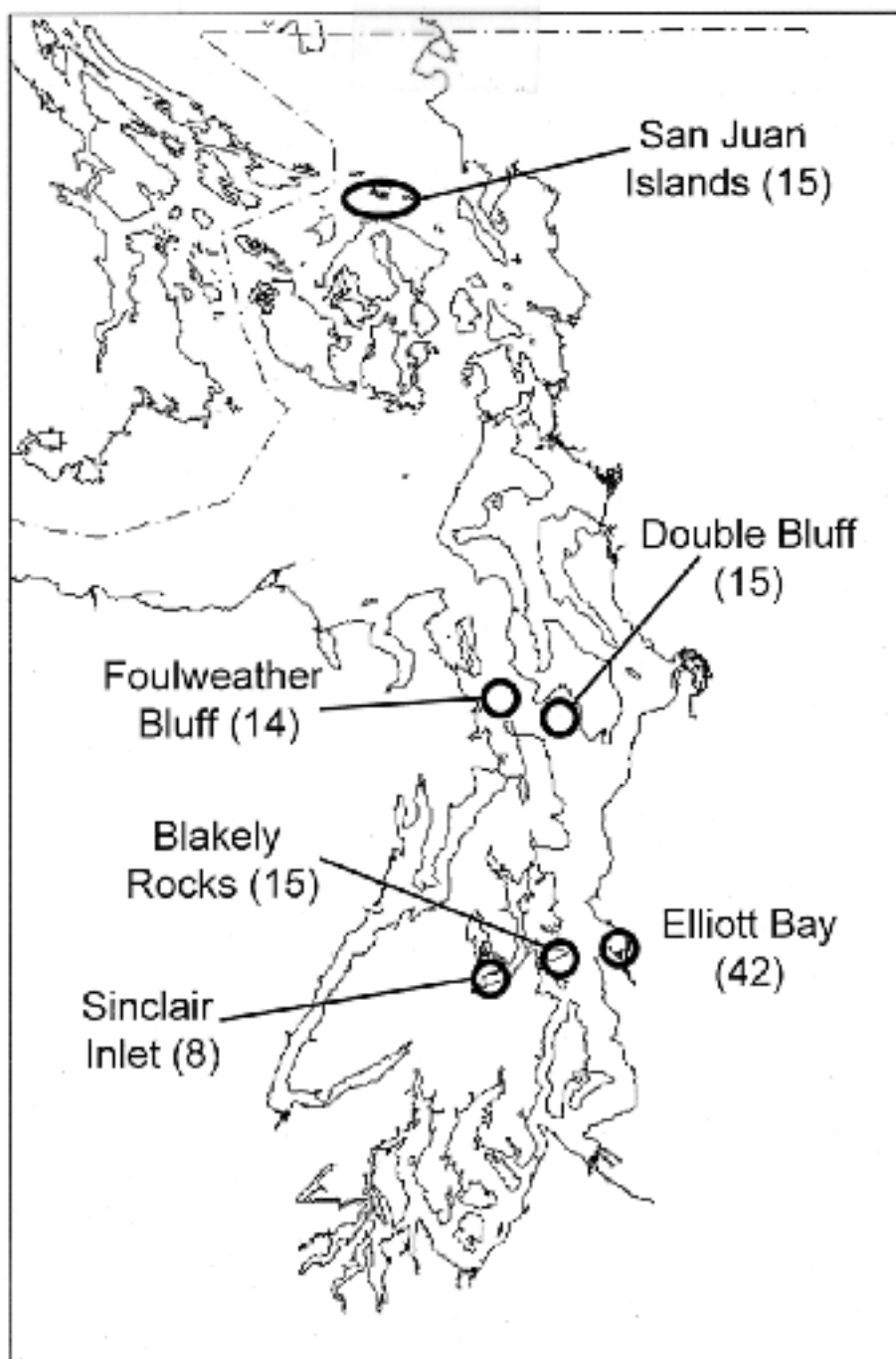


Figure 1. Location of sampling locations for quillback and brown rockfish in Puget Sound. Samples were taken from San Juan Islands, Double Bluff, and Blakely Rocks using hook-and-line in the fall of 1995; Foulweather Bluff and Sinclair Inlet were sampled using bottom trawls in the fall of 1997 and spring of 1996, respectively. Elliott Bay was sampled using hook-and-line and bottom trawls in the fall of 1995 and the spring of 1996 and 1997. Numbers in parentheses indicate sample sizes.

Total mercury concentration of individual muscle tissue samples was determined by cold-vapor atomic absorption after digestion with nitric/sulfuric (Puget Sound Estuary Program 1996a). PCBs were analyzed using gas-chromatography-electron capture detection with a dual megabore (Puget Sound

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Estuary Program 1996b). Tissue lipid concentration was estimated gravimetrically from samples where lipids were removed using acetone/methylene chloride extraction.

Least squares model I linear regression analysis was used to model the relationship between contaminant concentration and fish age. Stepwise forward linear regression variable selection with a confidence level of 95% was used to identify statistically significant factors. Location effects were isolated in regression analyses using dummy variables (Kleinbaum and Kupper 1978).

Table 1. Summary of fish age, length, tissue lipids, mercury and PCB data for quillback<sup>a</sup> and brown rockfish from six locations in Puget Sound, Washington.

Location		Fish Age (yrs)	Total Length (mm)	Tissue Lipids (%)	Mercury (mg/kg)	Total PCBs <sup>b</sup> (µg/kg)
Blakely Rocks	Mean	13.9	318	2.8	0.24	43.2
	Std. Dev.	5.2	20	4.0	0.11	34.0
	Min.	8	292	0.1	0.08	11.4
	Max.	28	373	15.8	0.46	138.8
	n	15	15	14	15	15
Double Bluff	Mean	12.6	319	0.4	0.22	5.1
	Std. Dev.	2.8	15	0.6	0.08	3.0
	Min.	7	297	0.1	0.10	4.0
	Max.	17	347	2.6	0.38	14.1
	n	15	15	15	15	15
Elliott Bay	Mean	13.9	308	1.1	0.38	122.4
	Std. Dev.	4.6	33	1.7	0.16	71.5
	Min.	5	220	0.1	0.08	54.4
	Max.	23	381	6.0	0.74	356.0
	n	42	39	15	39	19
Foulweather Bluff	Mean	7.5	321	---	0.29	---
	Std. Dev.	3.8	48	---	0.16	---
	Min.	3	233	---	0.14	---
	Max.	18	393	---	0.75	---
	n	14	14	---	14	---
San Juan Islands	Mean	16.0	363	0.5	0.26	3.9
	Std. Dev.	13.4	42	0.2	0.19	0.5
	Min.	6	292	0.2	0.08	2.0
	Max.	60	416	1.1	0.81	4.0
	n	15	15	14	15	15
Sinclair Inlet	Mean	24.3	372	0.4	0.84	268.3
	Std. Dev.	6.8	25	0.2	0.23	201.4
	Min.	14	340	0.1	0.51	84.7
	Max.	34	418	0.7	1.09	613.0
	n	8	8	8	8	8

<sup>a</sup> all samples were quillback rockfish except five brown rockfish from Sinclair Inlet

<sup>b</sup> sum of Aroclors 1254 and 1260

## Results

### Mercury

Concentrations of total mercury from individual fish ranged from 0.08 mg/kg in samples from Blakely Rocks, Double Bluff, Elliott Bay and San Juan Islands to 1.09 mg/kg from Sinclair Inlet (Table 1). Mean mercury concentration for four non-urban locations (Blakely Rocks, San Juan Islands, Foulweather Bluff, and Double Bluff) ranged from 0.22 to 0.29 mg/kg; mean mercury concentration from the two urban locations was 0.38 mg/kg (Elliott Bay) and 0.84 mg/kg (Sinclair Inlet). Mercury concentration was moderately to strongly correlated with fish age for all locations (linear regression analysis,  $r^2$  ranging from 0.48 to 0.82; Figure 2). Mean ages of rockfish varied widely among locations—from 7.4 years at Foulweather Bluff to 24.3 years from Sinclair Inlet. Thus, comparisons of mercury concentration among locations required an accounting of these location-specific age differences.

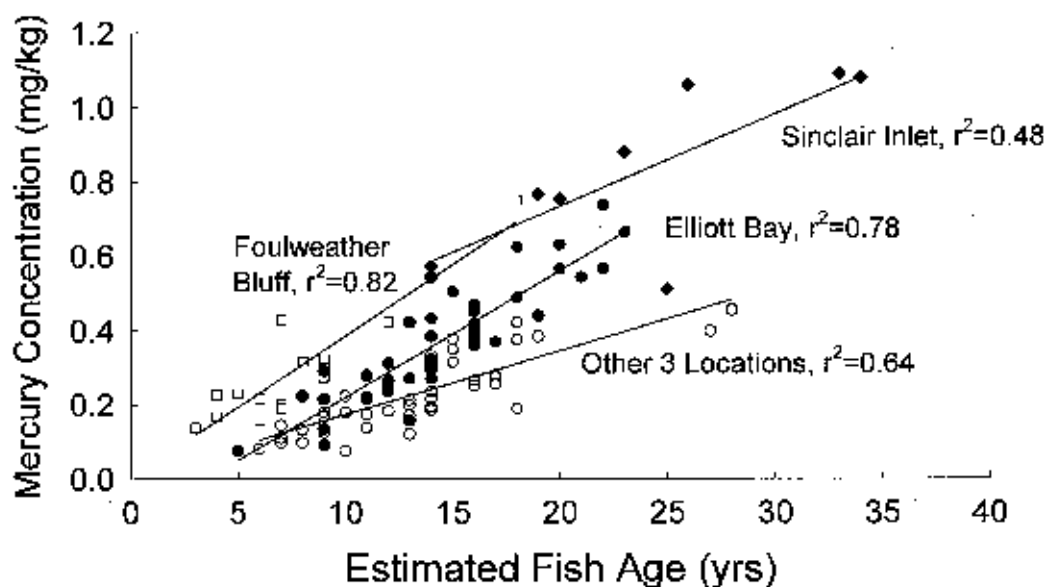


Figure 2. Accumulation of mercury in muscle tissue from quillback and brown rockfish sampled from Sinclair Inlet (diamonds), Foulweather Bluff (open squares), Elliott Bay (filled circles), and three locations (San Juan Islands, Double Bluff, and Blakely Rocks) grouped together (open circles). Lines were fitted using least squares model I linear regression analysis;  $p < 0.05$  for all regressions.

Comparing the relationships of mercury concentration with fish age graphically (Figure 2) allows an inspection and comparison of mercury concentration-at-age for each location. Mercury increased with age in rockfish from all locations; accumulation patterns from San Juan Islands, Double Bluff and Blakely Rocks were similar enough<sup>3</sup> to pool their data into a single linear regression (Figure 2). Sinclair Inlet, Elliott Bay, and Foulweather Bluff all had higher age-specific mercury concentration than the other three locations. We observed no differences in accumulation based on gender or tissue lipid levels.

Mercury in all eight samples of quillback and brown rockfish from Sinclair Inlet exceeded 0.5 mg/kg, and three exceeded 1.0 mg/kg; however, these were all relatively old fish, with ages ranging from 14 to 34 years. Rockfish from Foulweather Bluff appeared to have a similar pattern of accumulation as fish from Sinclair Inlet, but because there was little overlap in ages between the two locations, a valid comparison was not possible. Rockfish from Foulweather Bluff were all relatively young; their mean age was 7.5 years, compared to a range of means from 12.6 to 24.3 years for all the other locations. This resulted in a moderate mean mercury concentration for the location (0.29 mg/kg; Table 1). However,

for their respective age ranges, rockfish from Foulweather Bluff and Sinclair Inlet had the highest age-specific mercury concentrations.

Age-specific mercury concentration for Elliott Bay was intermediate, clearly higher than for the San Juan Islands/Double Bluff/Blakely Rocks group, and lower than for Sinclair Inlet or Foulweather Bluff<sup>d</sup>. The sample size for Elliott Bay was relatively large (39 individuals) which probably resulted in a more accurate description of the mercury concentration:age relationship than the other locations, however the oldest fish sampled from that location was only 23 years of age.

## **PCBs**

Polychlorinated biphenyls (PCBs), estimated as the sum of Aroclors 1254 and 1260, were consistently detected in three of five locations<sup>5</sup> (Table 1). Average PCB concentrations were 43.2, 122.4, and 268.3 µg/kg for Blakely Rocks, Elliott Bay, and Sinclair Inlet, respectively; Maximum PCB concentrations for these locations were 138.8, 356.0, and 613.0 µg/kg. PCBs were never detected in rockfish from San Juan Islands, and were detected (near the limit of detection) in only two of 15 samples in rockfish from Double Bluff.

We observed significant correlations between fish age and PCB concentration only in male rockfish from the two urban locations (Figures 3a and 3b). However, the regression for Sinclair Inlet was based on only five samples, and the Elliott Bay relationship appears to have been defined primarily by the two oldest samples, aged 20 and 22 years. Confidence in the validity of these correlations would be increased with more samples, especially in the range of ages greater than 20 years. PCBs did not accumulate in males from Blakely Rocks.

However, PCB concentrations in all males were low (<139 µg/kg) and the range of rockfish ages from that location was relatively low (eight to 18 years; Figure 3c). We observed no significant correlation ( $p>0.05$ ) between age and PCB concentration in female rockfish from any location (Figures 3a-c). In fact, PCB concentration seemed to decline with increasing age in female rockfish from Blakely Rocks (Figure 3c), however this weak correlation ( $r^2=0.24$ ) was not significant ( $p=0.12$ ).

Not shown is any analysis of PCBs and tissue lipid content. We inspected scattergrams for PCB concentration versus tissue-lipid content for each location-gender combination and observed no identifiable patterns.

## **Discussion**

Perhaps the most striking result we observed in this study is the strong, consistent pattern of accumulation of mercury in rockfish from all locations, with no effects attributable to gender or tissue-lipid levels. These accumulation patterns not only illustrated the relatively high mercury concentration in rockfish from contaminated areas that one might expect, but also showed that mercury accumulated in rockfish from uncontaminated areas. This presumed “natural” accumulation of normal or background mercury has been recognized in other marine organisms (Barber et al. 1972; Bernhard and Renzoni 1977; Barber 1984; Francesconi and Lenanton 1992; Monteiro et al. 1996; Phillips et al. 1997). Surprisingly however, we observed the greatest age-specific mercury concentration from Foulweather Bluff, a location that was selected as an uncontaminated reference area. Possible explanations for this are explored below.

In addition to age, strong location-specific factors other than gender and tissue lipid levels influenced the concentration of mercury in quillback and brown rockfish. The simplest explanation for these differences is related to variation in the source of mercury, where the concentration of mercury in the fish's tissue reflects regional levels of mercury. Strong associations of mercury concentration in sediments with resident organisms have been reported for a number of substrate-associated marine organisms world-wide (Kjørboe et al. 1983; Mikac and Picer 1985; Clark and Topping 1989; Leah et al. 1991; Palmer and Presley 1993; Collings et al. 1996; Herut et al. 1996). This association has also been observed in English sole from Puget Sound (Puget Sound Water Quality Action Team 1998).

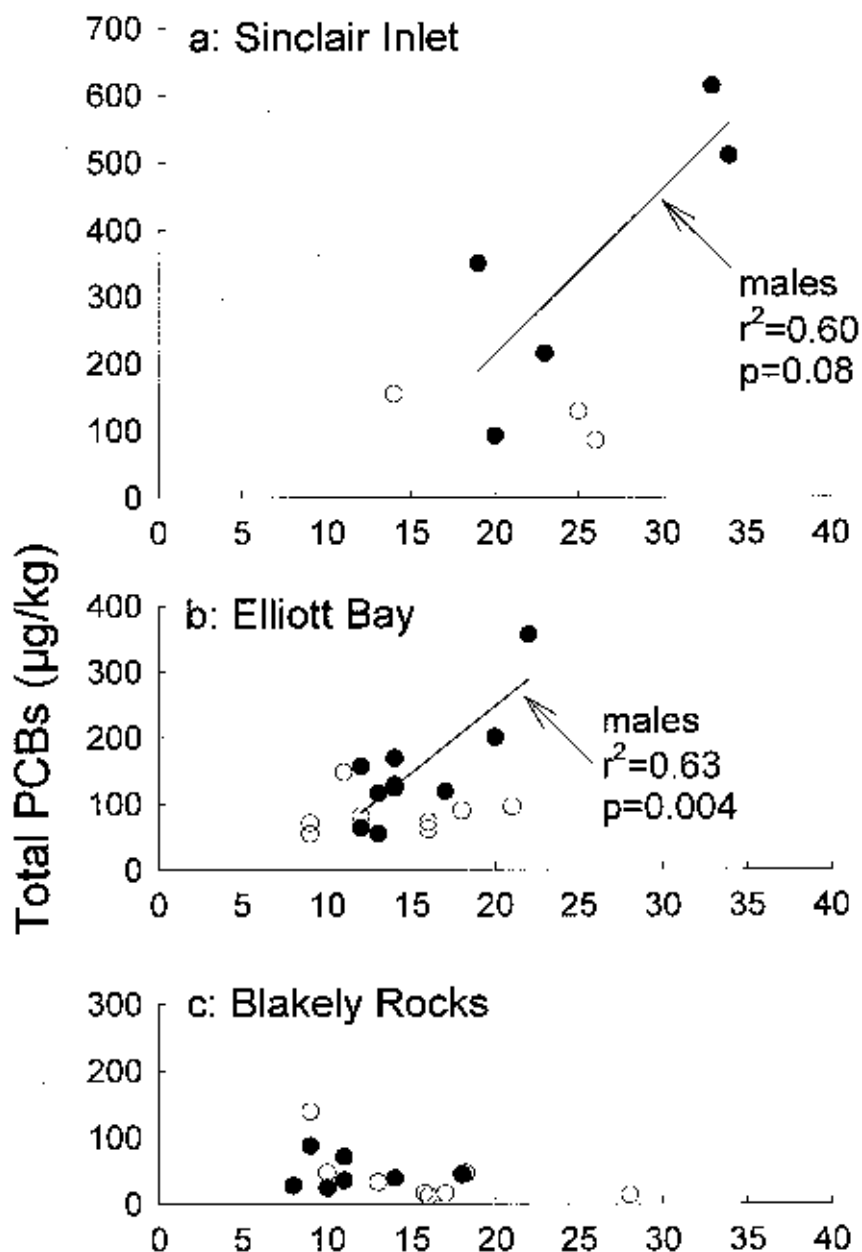


Figure 3. Concentration of PCBs and fish age for male (filled circles) and female (open circles) quillback and brown rockfish from three Puget Sound locations. Lines were fitted using least squares model I linear regression analysis;  $p=0.08$  and  $0.004$  for Sinclair Inlet and Elliott Bay regressions.

For rockfish in Puget Sound, PSAMP sediment chemistry data have shown that Sinclair Inlet and one location in Elliott Bay were contaminated with mercury (Puget Sound Water Quality Authority 1995), supporting the sediment-to-fish link. However, sediment samples taken in the 1970s from the Foulweather Bluff area had some of the lowest levels of mercury in the Puget Sound (Bothner 1973), some 30 to 50 times lower than Sinclair Inlet and Elliott Bay at that time. This suggests that some factor other than sediment contamination has caused a high concentration mercury in rockfish from Foulweather Bluff.

Consumption of contaminated prey, rather than absorption from the water, probably represents the primary pathway of mercury in marine fishes (Francesconi and Lenanton 1992), and the position a species occupies in the food web exerts a great influence on the level to which it biomagnifies persistent pollutants such as mercury (Young et al. 1980; Borgmann and Whittle 1992; Francesconi and Lenanton 1992; Hammar et al. 1993; Madenjian et al. 1993; Cabana et al. 1994; Madenjian et al. 1994; Davenport 1995; Kidwell et al. 1995; Stow 1995). These studies have demonstrated or inferred the importance of trophic level on the degree to which a species is exposed to persistent pollutants like mercury and PCBs; the result is that carnivorous organisms tend to accumulate these contaminants in greater concentrations than organisms lower in the food chain.

Because of the great diversity of prey consumed by rockfish, it is difficult to connect rockfish directly to sediment-associated contaminants via their prey. Quillback rockfish are carnivorous, consuming reef-associated prey, organisms from the finer-grained substrata that often surround reefs, and wide-ranging pelagic prey passing over reefs such as Pacific herring (Hueckel 1980; Murie 1994; Murie 1995). It is possible that rockfish from the Foulweather Bluff area consistently consumed organisms higher in the food chain than from other locations, resulting in a greater biomagnification of Hg.

Unlike mercury, PCBs appeared to accumulate only in male rockfish from Elliott Bay and Sinclair Inlet. These accumulation patterns are somewhat equivocal, because of the low sample size from Sinclair Inlet and lack of older specimens from Elliott Bay. In future sampling we will target older fish from urbanized locations like Sinclair Inlet and Elliott Bay to determine the validity of this pattern.

There is no evidence from our data that PCBs accumulated in female rockfish. Indeed, for females, we observed the greatest PCB concentrations in nine- and 10-year-old fish from Blakely Rocks and Elliott Bay, with lower concentrations in older fish. Larsson et al. (1996) hypothesized that for "lean" species which reproduce repeatedly throughout their lifetime (like rockfish), females should accumulate lipophilic compounds such as PCBs until they begin reproducing, after which contaminants are reduced via transfer of lipids to eggs (Figure 4a). Males in this hypothesis would continue to accumulate contaminants since there is no significant loss of PCB-rich lipids to sperm.

The age at which female quillback or brown rockfish in Puget Sound begin reproduction is unknown. (Wyllie-Echeverria 1987) estimated the age of first reproduction in quillback rockfish at five years for California populations. Our anecdotal observations of gonad conditions indicate that quillback rockfish from Puget Sound probably do not mature any earlier than five years, and probably mature substantially later. Additionally, rockfish fecundity increases exponentially with fish age (DeLacey et al. 1964; Washington et al. 1978), suggesting that reproductive loss of PCBs in females may increase with age once reproduction begins.

According to Larsson's (1996) hypothesis, lean species should exhibit a greater gender-specific disparity in PCB accumulation than fatty species. Rockfish are considered relatively "lean." Lipid content of muscle-tissue in rockfish from our study was usually less than 1%; we have recorded salmon lipid concentrations exceeding 20% (O'Neill and West, this volume). If Larsson's (1996) hypothesis is correct, we would expect to see accumulation of PCBs in female rockfish until sometime after age five, after which the intake of PCBs through diet would be offset by loss of PCBs through reproduction. Our data from Elliott Bay generally support this hypothesis (Figure 4b), however, the relationship is somewhat confused by the low PCB concentration observed in one 12-year-old specimen.

It is clear that age, gender, and location are important factors to consider when evaluating or predicting mercury or PCBs in long-lived species like rockfish. The life span of quillback and brown rockfish in Puget Sound is unknown. However, we have estimated ages up to 60 years for quillbacks, and 70- to 80-year-old rockfish are not uncommon in Alaska (Victoria O'Connell, Alaska Dept. Fish and Game, unpublished data). All of our regression analyses were performed on fish younger than 35 years, which indicates that we have underestimated the full range of mercury PCB concentrations in rockfish from Puget Sound.

The strong location-specific accumulation patterns indicate that Sound-wide predictions using a single fish age:contaminant regression model to predict mercury or PCBs may not be accurate. Gender

and location-specific factors such as sediment contamination and food habits may contribute significantly to the regional variability in contaminants we observed.

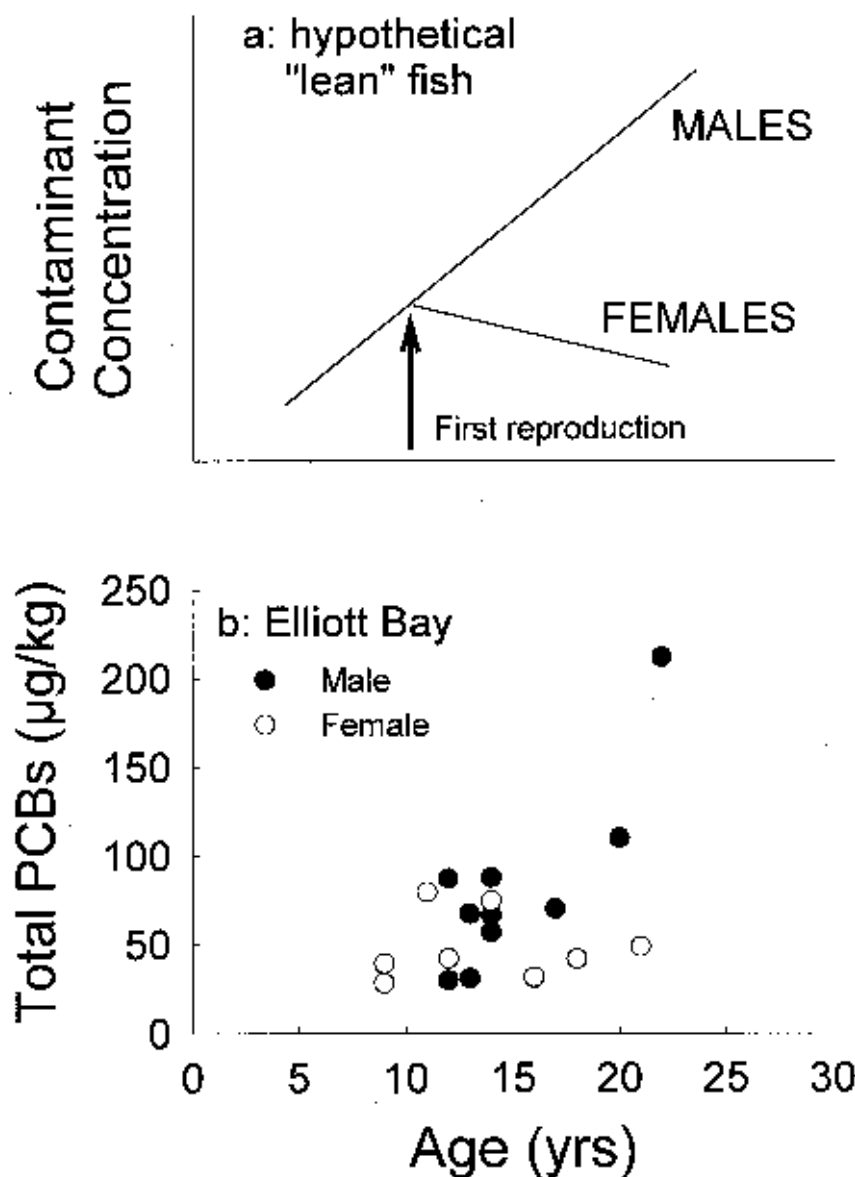


Figure 4. Hypothetical gender-specific accumulation patterns (a) of PCBs in "lean" fish that reproduce repeatedly throughout their lives (e.g., rockfish), adapted from Larsson et al. (1996), and gender-specific accumulation patterns (b) in quillback rockfish we observed from Elliott Bay, Washington.

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<sup>1</sup> We have estimated ages of quillback rockfish to 60 years in Puget Sound.

<sup>2</sup> We did not sample for organotins or dioxins.

<sup>3</sup> Stepwise linear regression analysis with dummy variables was used to separate location effects.

<sup>4</sup> Not shown in these analyses is the [Hg] for a single 60-year-old specimen taken from the San Juan Islands. That specimen had a [Hg] of 0.81 mg/kg and it was omitted from the regression analyses because of the large gap in data between it and the individual with the next lowest age (27 years).

<sup>5</sup> PCB results for Foulweather Bluff were not available as of this writing.